Validation of large-volume batch solar reactors for the treatment of rainwater in field trials in sub-Saharan Africa

Validation of large-scale solar reactors for the treatment of rainwater in field trials in sub-Saharan Africa

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Short title: Large-scale SODIS treatment of rainwater

Abbreviations[1]

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[1] ADWG – Australian drinking water guidelines; BDL – below detection limit; CFU – colony forming units; CPC – compound parabolic collector; DNA – deoxyribonucleic acid; DWAF – Department of Water Affairs and Forestry; *E. coli* – *Escherichia coli*; EMA – ethidium monoazide bromide; EU – European Union; FF – first-flush; HPC – heterotrophic plate count/heterotrophic bacteria; LB – luria bertani; PCA – principle component analysis; PET – polyethylene-terephthalate; PMA – propidium monoazide; PMMA – poly(methyl methacrylate); qPCR – quantitative polymerase chain reaction; RHRW – roof-harvested rainwater; ROS – reactive oxygen species; RWH – rainwater harvesting; SABS – South African Bureau of Standards; SODIS – solar disinfection; UV – ultraviolet radiation; WATERSPOUTT – Water Sustainable Point-Of-Use Treatment Technologies; WHO – World Health Organisation; WSP – water safety plan; Zn – zinc.

Abstract

The efficiency of two large-scale solar reactors [Prototype I (140 L) and II (88 L)] in treating rainwater on-site in a local informal settlement ~~(Site 1)~~ and farming community ~~(Site 2)~~ was assessed. Untreated ~~(Tank 1 and Tank 2 FF)~~ and treated (Prototype I and II) tank water samples were routinely collected from each site and all the measured physico-chemical parameters, anions and cations were within national and international drinking water guidelines limits. Culture-based analysis indicated that *Escherichia coli*, total and faecal coliforms, enterococci and heterotrophic bacteria counts exceeded drinking water guideline limits in 61%, 100%, 45%, 24% and 100% of the untreated tank water samples collected from both sites. However, an 8 hour solar exposure treatment for both solar reactors was sufficient to reduce these indicator organisms to within drinking water standards, with the exception of the heterotrophic bacteria which exceeded the drinking water guideline limit in 43% of the samples treated with the Prototype I reactor (1.01 log reduction). Molecular viability analysis subsequently indicated that mean overall reductions of 75% and 74% were obtained for the analysed indicator organisms (*E. coli* and enterococci) and opportunistic pathogens (*Klebsiella*, *Legionella*, *Pseudomonas*, *Salmonella* and *Cryptosporidium* oocysts) in the Prototype I and II solar reactors, respectively. The large-scale solar reactor prototypes could thus effectively provide three (88 L Prototype II) to five (144 L Prototype I) people on a daily basis with the basic water requirement for human activities (25 L). Additionally, the outlined water safety plan may aid in identifying how and where rainwater harvesting systems should be installed and maintained to ensure the quality of the treated water.

Keywords: Rainwater harvesting; solar disinfection; rainwater quality; sub-Saharan Africa

1. Introduction

The Global Risks Report released for 2019 listed water crises as one of the top ten risks in terms of likelihood (rating of 9; very likely to occur) and impact (rating of 4; severe impact) (Global Risks Report, 2019). The probability of a water crisis risk in sub-Saharan Africa is significantly increased as a high proportion of the population reside in urban informal settlements and rural areas, with limited access to a safe water supply and sanitation infrastructure (Dos Santos et al. 2017). However, as highlighted by Gwenzi and Nyamadzawo (2014) and Emenike et al. (2017), rainwater is considered an under-exploited water source in sub-Saharan Africa and may serve as an effective reserve to improve and encourage equity in water access. Roof-harvested rainwater (RHRW) can however, be contaminated with various chemicals and microorganisms, which may limit its use as a potable water source (Hamilton et al. 2019). While the chemical pollutants have not been directly associated with the incidence of disease, organic debris and faecal matter from animals and birds that have access to the catchment surface, have been identified as the primary sources of microbial contaminants such as *Legionella*, *Klebsiella*, *Pseudomonas* and *Cryptosporidium* (Hamilton et al. 2019).

Treatment strategies that may be implemented to improve the quality of rainwater include the utilisation of gutter screens or first-flush diverters for the prevention of contaminant entry into the collection tank or post-collection treatment [chemical (e.g. chlorination) and physical treatments (e.g. filtration, solar disinfection (SODIS) and thermal disinfection)] (Hamilton et al. 2019). Although various chemical and physical treatment technologies have been investigated, SODIS is considered a cost-effective treatment method and is recommended by the World Health Organisation (WHO) for the effective reduction of microbial contamination in water sources (Ubomba-Jaswa et al. 2010). In its simplest form, SODIS entails filling a transparent container [usually a 2 L ~~or 5 L~~ polyethylene-terephthalate (PET) bottle] with contaminated water and exposing the bottle to direct sunlight for six to eight hours to allow ultraviolet (UV) radiation and solar-mild heat to inactivate microbial contaminants

(McGuigan et al. 2012). Ultraviolet radiation directly inactivates the microbial contaminants by damaging nucleic acids and leads to the formation of reactive oxygen species (ROS), which react and damage proteins, nucleic acids and membrane lipids (Nelson et al. 2018). The water temperature will also increase as water molecules absorb the UV radiation, which leads to cell membrane damage. The major drawbacks associated with this technique are however, the small volumes of water that can effectively be treated (2 to 5 L) and decreased efficiency during overcast weather conditions (up to 48 hours of treatment). Increases in treatment volume and efficiency may then be obtained by employing various modifications (SODIS enhancement technologies) such as solar mirrors (concentrates UV radiation) and larger reactor tubes (increase treatment volume) (Ubomba-Jaswa et al. 2010; McGuigan et al. 2012).

As part of the European Union (EU) Horizon 2020 project titled Water Sustainable Point of Use Treatment Technologies (WATERSPOUTT), Polo-López et al. (2019a) investigated various enhancement technologies that may cost-effectively allow for larger volumes of water to be treated using SODIS. Results from the study indicated that the use of a static batch reactor system employing ~~U-type solar mirrors~~ allowed for the effective treatment of a larger volume (68% more) of water as compared to the compound parabolic collector (CPC)-type solar mirrors under the same solar exposure conditions (Polo-López et al. 2019a). In a follow-up study, the same research group designed two large-scale solar reactor prototypes (static batch systems with 88 L and 140 L treatment volumes, respectively), where multiple poly(methyl methacrylate) (PMMA) reactor tubes were positioned in the centre of U-type solar mirrors (Polo-López et al. 2019b). Preliminary assessment of the solar reactor prototypes, using spiked synthetic rainwater samples and culture-based analysis, indicated that a ≥ ~~6~~ log removal efficiency was obtained for *Escherichia coli* (*E. coli*) and *Salmonella enteriditis* after 1.5 hour natural sunlight exposure, while a 2 hour sunlight exposure was required to achieve the same log reduction for *Enterococcus faecalis* and *Pseudomonas aeruginosa* (*P. aeruginosa*).

The primary aim of the current study was to assess the efficiency of the two newly designed large-scale solar reactor prototypes (Polo-López et al. 2019b) for the treatment of

RHRW on-site in a local informal settlement (140 L Prototype I) and a rural farming community (88 L Prototype II). The chemical quality of the RHRW before and after solar reactor treatment was routinely assessed by monitoring various physico-chemical parameters (e.g. temperature, pH, and turbidity), anions and cations. Additionally, the removal of traditional indicator organisms (*E. coli*, total and faecal coliforms, enterococci and heterotrophic bacteria) and selected opportunistic pathogens (*Klebsiella* spp., *Pseudomonas* spp. and *Salmonella* spp.), was assessed using culture-based analysis. Ethidium monoazide bromide quantitative polymerase chain reaction (EMA-qPCR) assays were also used to monitor the reduction efficiency of indicator organisms (*E. coli* and enterococci) and opportunistic pathogens (*Klebsiella* spp., *Legionella* spp., *Pseudomonas* spp., and *Salmonella* spp.), while propidium monoazide (PMA) qPCR assays were used to monitor *Cryptosporidium* oocyst reductions. A water safety plan (WSP) outlining guidelines for the use of rainwater harvesting combined with solar reactor treatment was also implemented.

2. Materials and methods

2.1 Description of large-scale solar reactor prototypes and sampling sites

Two large-scale solar reactor prototypes were designed and constructed as part of Work Package 1 (WP1) by the WATERSPOUTT research consortium as part of a EU Horizon 2020 project under grant agreement no. 688928 for implementation in South Africa and Uganda. Detailed information on the design and working mechanisms of the systems are outlined in Polo-López et al. (2019b), with the current study focussing on the application of these systems in field trials in South Africa. The Prototype I solar reactor (140 L treatment volume) was installed in Enkanini informal settlement (Site 1; GPS coordinates: 33°55'28.1"S 18°50'35.8"E) during July 2018 and consisted of three PMMA reactor tubes (200 mm diameter) that were positioned in the centre of a U-type solar mirror (constructed from anodized aluminium). The reactor tubes were positioned at a 34° angle (equal to the local latitude) and were inter-connected by UV-A transparent PMMA tubing (Fig. 1A). The Prototype II solar reactor (88 L

127 treatment volume) was installed next to a local church building in the Skoolplaas farming
128 community (Site 2; GPS coordinates: 33°56'38.5"S 18°46'26.3"E) during July 2018 and
129 consisted of the same materials and design as Prototype I, with the exception that eight PMMA
130 tubes (100 mm diameter) were used in the system (Fig. 1B). Additionally, as space was
131 available between the gutter system and the rainwater harvesting (RWH) tank at site 2, a first-
132 flush (FF) diverter (Superhead® rainwater filter) was installed to redirect the initial roof run-off
133 during a rain event (Fig. 1B). A detailed description of the sampling sites and system
134 installation is outlined in Appendix A.

135 **2.2 Sample collection**

136 For the microbial and chemical analysis of the water produced by the solar reactor prototypes
137 (Fig. 1), an untreated 10 L sample was collected directly from the RWH tank at each site
138 [hereafter referred to as Tank 1 (Site 1) and Tank 2-FF (Site 2)]. The respective solar reactor
139 prototypes were filled with tank water from the RWH tanks and exposed to direct sunlight for
140 6 hours (sampling sessions 1 to 8) or 8 hours (sampling sessions 9 to 18). Following the solar
141 exposure, 10 L of each treated sample was collected directly from the solar reactor ~~prototypes~~
142 [hereafter referred to as Prototype I (Site 1) and Prototype II (Site 2)]. Based on the availability
143 of rainwater in the RWH tanks, 15 sampling sessions were conducted at site 1 (n = 30; August
144 2018 to March 2019), while 18 sampling sessions were conducted at site 2 (n = 36; August
145 2018 to April 2019). For ease of presentation, sampling sessions 1 to 18 are designated as
146 #1 (sampling session 1), #2 (sampling session 2), etc., throughout the manuscript.

147 The temperature, pH and total dissolved solids present in all water samples were
148 measured using a hand-held Milwaukee Instruments MI806 meter (Spraytech, South Africa),
149 while the dissolved oxygen was measured using a Milwaukee Instruments M600 meter
150 (Spraytech, South Africa). Rainfall and daily ambient temperature data for the study period
151 was obtained from the South African Weather Services, while solar irradiance data [mean
152 ambient UV-A and UV-B radiation] was obtained from the Stellenbosch Weather Services
153 [Stellenbosch University, Faculty of Engineering (http:// weather.sun.ac.za/)].

2.3 Chemical analysis

The chemical quality of the untreated and solar reactor treated tank water samples was determined by monitoring cation and anion concentrations and turbidity as described by Strauss et al. (2018). All samples (*n* = 66) were monitored for cations, while representative samples (*n* = 22; #1, #7, #10, #12, #15 and #18) were monitored for anions and turbidity.

2.4 Culturing of indicator organisms and opportunistic pathogens

The microbial quality of the tank water samples collected from sites 1 and 2 were monitored before (untreated) and after solar reactor treatment using various culture-based analyses. *Escherichia coli* and total coliforms were enumerated simultaneously using membrane filtration as described by Dobrowsky et al. (2015), while enterococci, faecal coliforms and the heterotrophic plate count/bacteria (HPC) were enumerated as outlined in Strauss et al. (2016), with a minor modification; Luria Bertani (LB) agar (Biolab, Merck, South Africa) replaced Reasoner's 2A agar (Oxoid, Hampshire, England) for the enumeration of HPC. For the treated samples (Prototypes I and II) where the HPC were reduced to below the detection limit [BDL; < 1 colony forming units (CFU)/1 mL], the potential regrowth of bacteria was monitored. Briefly, 20 mL of each treated sample was stored in a sterile McCartney bottle at room temperature and 100 µL of the treated water was spread plated onto LB agar (Biolab, Merck) every 24 hours for a period of 2 days. The plates were then incubated at 37 °C. Additionally, *Klebsiella* spp., *Pseudomonas* spp. and *Salmonella* spp. were enumerated as outlined in Clements et al. (2019), while coliphages were enumerated as outlined by Baker et al. (2003) using *E. coli* ATCC 13706 as the target bacterial host. All culture-based analyses were performed in duplicate.

2.5 Tank water concentration, viability treatment and DNA extraction

The concentration of 1 L (Site 1) and 2 L (Site 2) samples, EMA treatment and subsequent DNA extractions were performed for each of the samples collected before and after solar

reactor treatment as outlined in Reyneke et al. (2016). For the molecular quantification of *Cryptosporidium* spp. within the collected samples, the same methodology was repeated with the exception that a PMA treatment as described by Alonso et al. (2014) was followed.

2.6 Molecular-based enumeration of indicator organisms and opportunistic pathogens

Quantitative PCR was performed in order to quantify *E. coli*, enterococci, *Klebsiella* spp., *Legionella* spp., *Pseudomonas* spp. and *Salmonella* spp. in all of the collected tank water samples, while *Cryptosporidium* oocysts were quantified in the samples collected from #9 to #15 and #9 to #18 for sites 1 and 2, respectively. All qPCR assays were conducted using a LightCycler® 96 (Roche Diagnostics, Risch-Rotkreuz, Switzerland) instrument in combination with the FastStart Essential DNA Green Master Mix (Roche Diagnostics) as outlined in Reyneke et al. (2017), with the primer pairs and cycling parameters presented in Table A1. Standard curves for the respective qPCR assays were generated using the methodology outlined in Reyneke et al. (2017), while the qPCR performance characteristics of the various assays were analysed using the Roche LightCycler® 96 Software Version 1.1. Furthermore, to compensate for the different sample volumes used per site for rainwater concentration [1 L (Site 1) and 2 L (Site 2)] the gene copies detected in the samples utilising the qPCR assays were converted to gene copies per 100 mL of the original tank water sample as outlined by Waso et al. (2018). The gene copy numbers (gene copies/100 mL) were then converted to cell equivalents (cells or oocysts/100 mL) by utilising the number of copies of the target gene present within the target host (Table A1). All final concentrations for qPCR analyses are thus presented as equivalent cells or oocysts/100 mL original tank water sample.

2.7 Maintenance of prototype reactors and water safety plan

Following the system installations, workshops were conducted within the respective communities to outline the principle of rainwater harvesting, the working mechanism and operational maintenance of the solar reactors. Information on the domestic activities (i.e. laundry, cleaning, washing, etc.) the treated rainwater could be used for was also provided

(Fig. A3). Exemption from ethical clearance was obtained from the Research Ethics Committee (Humanities) Stellenbosch University (Ethics Reference no.: SU-HSD-004624), as the participating households would not be using the treated water for drinking purposes.

As outlined by the WHO (2004), the most efficient way of consistently ensuring the safety of a drinking water supply is through the utilisation of a WSP (Appendix B), which may be defined as a risk assessment and management approach that monitors the entire water supply process (e.g. collection of RHRW to utilisation of treated tank water by the consumer). The first step in the development of the WSP was to identify all potential hazards/hazardous events that may influence the quality of rainwater during the harvesting process, storage and treatment process (Appendix B), using published literature and personal observations at the respective study sites, during the study period. Additionally, various maintenance and remedial actions were identified to prevent certain water safety hazards (e.g. prevent organic debris from entering the storage tank) or to implement after a hazardous event occurred (e.g. control measure failed and organic debris washed into the storage tank) (Appendix B). Following the identification of the potential hazards, a risk assessment matrix (Appendix C) was compiled that would enable the risk characterisation associated with each hazard/hazardous event and enable the assessment of the various control measures (e.g. maintenance strategies, use of a first-flush diverter system etc.) in eliminating the identified water safety hazards.

2.8 Statistical analysis

Statistical analyses were conducted utilising either RStudio (version 1.0.153) or Microsoft Excel® Ver. 15.31. Overall differences in sample composition between site 1 and site 2 and the untreated (Tank 1 and Tank 2) and solar reactor treated (Prototype I and II) tank water samples was determined by evaluating all measured physico-chemical, chemical and microbial parameters using the parametric paired *t*-test (significant when $p < 0.05$). Principle component analysis (PCA) was then used to visualise the correlations between the measured

231 cations at both sites and identify which cations primarily influenced the sample composition at
232 each site.

233 **3. Results and Discussion**

234 **3.1 Physico-chemical properties and chemical analysis of the collected tank water**
235 **samples**

236 The mean ambient UV-A radiation at both sampling sites ranged from 7.16 W/m^2 (12/09/2018)
237 to 31.29 W/m^2 (14/01/2019), while the mean ambient UV-B radiation ranged from 1.33 W/m^2
238 (12/09/2018) to 4.63 W/m^2 (14/01/2019) (Table A2). The untreated tank water temperature at
239 site 1 (Tank 1) ranged from 9.0 °C (02/08/2018 and 15/08/2018) to 24.0 °C (28/01/2019), with
240 a mean temperature of 16.3 °C recorded for all sampling days, while the tank water
241 temperature in the samples collected from the Prototype I solar reactor ranged from 15.5 °C
242 (12/09/2018) to 45.0 °C (28/01/2019) (mean 28.9 °C). Similarly, the untreated tank water
243 temperature at site 2 (Tank 2-FF) ranged from 10.0 °C (15/08/2018) to 26.0 °C (25/10/2018)
244 (mean 18.1 °C), while the tank water temperature in the samples collected from the Prototype
245 II solar reactor ranged from 18.0 °C (12/09/2018) to 46.5 °C (28/01/2019) (mean 32.6 °C).

246 All measured physico-chemical parameters (pH, turbidity, electrical conductivity, total
247 dissolved solids and dissolved oxygen) in the collected untreated and prototype treated
248 rainwater samples adhered to the drinking water guideline limits of the South African
249 Department of Water Affairs and Forestry (DWAF) (DWAF, 1996), South African National
250 Standards (SANS) 241 [South African Bureau of Standards (SABS), 2005], Australian
251 Drinking Water Guidelines (ADWG) (NHMRC and NRMMC, 2011) and WHO (2011), with no
252 significant difference ($p > 0.05$) observed for the data collected for the untreated and treated
253 (Tank 1 and Prototype I; Tank 2-FF and Prototype II) tank water samples or between sites 1
254 and 2 (Tank 1 and 2-FF) (Table A3).

255 Results for the chemical analyses of the untreated (Tank 1 and Tank 2-FF) and treated
256 (Prototype I and Prototype II) tank water samples collected from sites 1 and 2, indicated that

all anions and cations (Table A3) were within the respective drinking water guideline limits [DWAF, 1996; SANS 241 (SABS, 2005); ADWG (NHMRC and NRMMC, 2011); WHO, 2011], with the exception of the mean zinc (Zn) concentration recorded in the samples collected from site 1 [Tank 1 (mean of 3044 µg/L) and Prototype I (mean of 3061 µg/L)]; which exceeded (albeit not significantly) the DWAF (1996) and ADWG (NHMRC and NRMMC, 2011) limit of 3000 µg/L. However, these samples were within the 5000 µg/L SANS 241 (SABS, 2005) limit. The increased Zn concentrations recorded at site 1 (Tank 1 and Prototype I), in comparison to site 2 (Tank 2-FF and Prototype II), may primarily be attributed to the metal sheeting (e.g. Zn sheeting) roofing material used to construct the catchment system, as the leaching of metals from metal roofing materials (corrosion during rain events and continuous exposure to sunlight) have been reported to be a major contributor of metal ions in rainwater (Chang et al. 2004; Reyneke et al. 2018). It should be noted, that while the catchment system at site 2 was also constructed from Zn sheeting roofing material, the entire surface of the catchment system was painted with a weather resistant roof paint (personal communication) which may have limited the leaching of metal ions into the rainwater. Additionally, the first-flush diverter connected to the rainwater tank at site 2 (Tank 2-FF) may have improved the physico-chemical quality of the tank water samples. First-flush diverter systems act as a pre-treatment barrier by redirecting the initial roof run-off water (at the start of a rain event), which is thought to contain the highest concentration of pollutants (Sánchez et al. 2015). Gikas and Tsihrintzis (2012) compared the quality of RHRW collected in the flush pipe of first-flush diverter systems, with the RHRW entering the collection tanks (RWH tanks) and reported that all measured mean anion and cation concentrations were higher in the collected first-flush samples. The authors concluded that the diversion of the first-flush roof run-off away from the collection tanks may improve the physico-chemical quality of the RHRW.

As no significant difference was obtained when comparing the anion and cation concentrations (Table A3) recorded in the untreated tank water samples to the treated tank water samples (Tank 1 vs Prototype I, Tank 2-FF vs Prototype II) and the tank water samples from each site clustered together (Fig. 2), it was concluded that the solar reactor prototypes

285 (system components and the treatment mechanism) did not influence the chemical quality of
286 the tank water samples.

287 **3.2 Removal efficiency of indicator bacteria and opportunistic pathogens**

288 **3.2.1 Culture-based analysis**

289 For the untreated tank water samples collected from site 1 (Tank 1; *n* = 15), the *E. coli*, faecal
290 coliform, total coliform, enterococci and HPC concentrations exceeded the respective drinking
291 water guideline limits in 67%, 73%, 100%, 20% and 100% of the samples, respectively (Table
292 1). Analysis of the corresponding treated samples (Prototype I; *n* = 15) indicated that the
293 *E. coli* (> 0.78 log reduction), enterococci (> 3.48 log reduction) and faecal coliform (> 4.08
294 log reduction) concentrations were reduced to BDL (< 1 CFU/100 mL) in all the collected
295 samples. Total coliforms were reduced to BDL in 63% of the treated samples collected
296 following a 6 hour solar exposure (# 1-8) (> 3.94 log reduction), with a mean of 55 CFU/100 mL
297 detected in the samples (37%) where total coliform counts above the standard were detected.
298 An increase in solar exposure to 8 hours (# 9-15) resulted in an increased treatment efficiency,
299 as total coliforms were reduced to within the 5 CFU/100 mL DWAF (1996) and
300 10 CFU/100 mL SANS 241 (SABS, 2005) guideline limits in 100% of the treated samples (4.66
301 log reduction). For the HPC analysis, 38% of the treated samples were reduced to within the
302 drinking water guideline limit of 1.0×10^4 CFU/100 mL (1.71 log reduction) after a 6 hour solar
303 exposure [mean of 2.4×10^4 CFU/100 mL detected in the remaining 63% samples (1.21 log
304 reduction)], while 57% of the treated samples were reduced to within the guideline limit (2.08
305 log reduction) after an 8 hour solar exposure [mean of 2.7×10^4 CFU/100 mL detected in the
306 remaining 43% of samples (1.01 log reduction)] (Fig. A6).

307 For the untreated tank water samples collected from site 2 (Tank 2-FF; *n* = 18), the
308 *E. coli*, faecal coliform, total coliform, enterococci and HPC concentrations exceeded the
309 respective drinking water guideline limits in 56%, 22%, 100%, 28% and 100% of the samples,
310 respectively (Table 1). Analysis of the corresponding treated samples (Prototype II; *n* = 18)

311 indicated that the *E. coli* (> 0.48 log reduction), enterococci (> 3.34 log reduction) and faecal
312 coliform (> 3.04 log reduction) concentrations were reduced to BDL (< 1 CFU/100 mL) in all
313 collected samples, while total coliforms were reduced to within the 5 CFU/100 mL DWAF
314 (1996) and 10 CFU/100 mL SANS 241 (SABS, 2005) guideline limits (3.85 log reduction).
315 Heterotrophic bacteria were then reduced to within the 1.0×10^4 CFU/100 mL DWAF (1996)
316 drinking water guideline limit in 88% of the treated samples (mean of 4.6×10^3 CFU/100 mL
317 recorded) after a 6 hour solar exposure (# 1-8) (2.11 log reduction), with a mean of
318 1.8×10^4 CFU/100 mL detected in the samples (12%) where HPC concentrations above the
319 standard were detected. In comparison, 100% of the treated samples were reduced to within
320 the 1.0×10^4 CFU/100 mL drinking water guideline limit after an 8 hour solar exposure (# 9-
321 18) (≥ 2.02 log reduction; Fig. A6).

322 *Klebsiella* spp. were detected in 100% (mean concentration of 1.9×10^4 CFU/100 mL)
323 and *Salmonella* spp. in 60% (mean concentration of 6.3×10^3 CFU/100 mL) of the untreated
324 rainwater samples collected from site 1 (Tank 1); however, both organisms were reduced to
325 BDL (> 4.28 and > 3.8 log reduction, respectively) following treatment using the Prototype I
326 solar reactor (Table 1). *Klebsiella* spp. were also detected in 17% (mean concentration of
327 8.0×10^2 CFU/100 mL) and *Salmonella* spp. in 6% (mean concentration of
328 1.0×10^3 CFU/100 mL) of the untreated rainwater samples collected from site 2 (Tank 2-FF),
329 with both organisms reduced to BDL (> 2.9 and > 3 log reduction, respectively) following
330 treatment using the Prototype II solar reactor (Table 1). *Pseudomonas* spp. and coliphages
331 were not detected in any of the rainwater samples collected from sites 1 and 2.

332 Although numerous studies have investigated the use of SODIS to treat contaminated
333 water, varying degrees of treatment efficiency (0.46 to > 6 log reductions in bacteria) have
334 been reported depending on experimental design (McGuigan et al. 2012; Hamilton et al.
335 2019). However, a limitation of SODIS which has consistently been highlighted by these
336 investigators is the small treatment volume (2 to 5 L). Ubomba-Jaswa et al. (2010) investigated
337 the use of a 25 L SODIS reactor (methacrylate tube) situated inside a CPC and reported on
338 the complete inactivation of *E. coli*, even during unfavourable weather conditions (cloudy with

low solar intensity). Polo- López et al. (2019a) then expanded on this research and investigated cost-effective SODIS enhancement strategies that would enable the treatment of larger volumes of water (32 L and 54 L), with the results obtained leading to the design of the large-scale solar reactor prototypes (Prototype I and II) assessed in the current study. The treatment efficiency of the Prototype I and II solar reactors was also assessed by Polo-López et al. (2019b) under controlled conditions, by spiking synthetic rainwater with laboratory strains of *E. coli*, enterococci, *Salmonella* and *Pseudomonas* (10^5 – 10^6 CFU/mL bacterial cells) using a 6 hour solar exposure treatment time. A ≥ 6 log reduction of all the test bacteria was obtained, with the system classified as "highly protective (≥ 4 log reduction)" against bacteria according to the WHO (2016) household water treatment technology performance criteria. In comparison, results from the current study, for both solar reactor prototypes, during a 6 hour solar exposure treatment, indicated that ≥ 2.54 log reduction was obtained when monitoring the removal of enterococci, faecal and total coliforms, while mean log reductions of ≥ 1.21 log were obtained for the removal of HPC. Based on these results, the 6 hour solar exposure treatment with the prototypes in field trials failed to meet the ≥ 2 log removal required for a "protective" classification against bacteria. The Polo-López et al. (2019b) study was however, conducted in a hot arid climate (Tabernas Dessert, Almería, Spain) with a mean UV radiation of 28.31 W/m^2/h recorded during the 6 hour treatment trials, while the field trials of the systems in the current study were conducted in a moderate Mediterranean climate (Stellenbosch, Western Cape, South Africa), where a mean UV radiation of 20.82 W/m^2/h was recorded during the 6 hour treatment trials (Table A2).

The treatment time in the current study was subsequently increased to 8 hours (Site 1: #9-15; Site 2: #9-18) in order to increase the overall UV dose (mean UV radiation of 24.72 W/m^2/h was recorded from #9-18). For both prototypes a ≥ 3.44 log reduction was subsequently obtained when monitoring the removal of enterococci, faecal and total coliforms, while the mean log reductions for the removal of HPC increased to ≥ 2.02 log. Based on the observed treatment efficiencies obtained using the Prototype I and II solar reactors in the current study (8 hour treatment), the prototypes may be classified as "protective (≥ 2 log

reduction)", for the removal of bacteria in the tank water (WHO, 2016). More importantly, culture-based analysis indicated that both treatment systems were able to produce water that adhered to the microbial parameters as stipulated in the respective drinking water guidelines [DWAF, 1996; SANS 241 (SABS, 2005); ADWG (NHMRC and NRMMC, 2011); WHO, 2011], with lower indicator organism counts recorded in the tank water samples collected from site 2, where the first-flush diverter system was installed. The treated water collected from the large-scale solar reactor prototypes could however, only be stored for a maximum of 24 hours, as microbial re-growth occurred after this point.

3.2.2 Molecular-based analysis

The performance characteristics of the respective qPCR assays are provided in Table A4. Results obtained using EMA-qPCR indicated that an overall mean decrease of 83.76% (0.79 log reduction) in intact *E. coli* cells was recorded after treatment using Prototype I, while an overall mean decrease of 82.76% (0.76 log reduction) was recorded after treatment for Prototype II (Fig. 3). Similarly, intact enterococci cells decreased by a mean of 91.68% (1.08 log reduction) after treatment using Prototype I, while an 84.89% (0.82 log reduction) mean decrease was recorded after treatment using Prototype II (Fig. 3). In comparison, quantification of intact *Klebsiella* cells indicated that this genus was more resistant to the solar reactor treatment as mean decreases of 62.44% (0.43 log reduction) and 60.42% (0.40 log reduction) were recorded after treatment using Prototype I and II, respectively (Fig. 3). Similarly, intact *Legionella* cells decreased by 68.61% (0.50 log reduction) after treatment using Prototype I and by 63.77% (0.44 log reduction) after treatment using Prototype II (Fig. 3). Overall mean decreases in intact *Pseudomonas* cells of 79.09% (0.68 log reduction) and 87.50% (0.90 log reduction) were recorded after treatment using Prototype I and II, respectively, while *Salmonella* cells decreased by 78.36% (0.66 log reduction) after treatment using Prototype I and 67.82% (0.49 log reduction) after treatment with Prototype II (Fig. 3). Lastly, PMA-qPCR analysis indicated that *Cryptosporidium* oocysts decreased by 57.14%

(0.62 log reduction) after treatment using Prototype I, while a mean decrease of 73.81% (0.58 log reduction) was recorded after treatment using Prototype II (Fig. 3).

Overall, the EMA-qPCR and PMA-qPCR analysis indicated that the Prototype I and II solar reactors reduced the opportunistic pathogens by 74.43%. This discrepancy in the observed treatment efficiency in comparison to the results obtained using culture-based analysis, may be attributed to EMA-qPCR and PMA-qPCR detecting viable but non culturable (VBNC) cells within the water samples (Fittipaldi et al. 2012; Mansi et al. 2014). It has been reported that certain opportunistic pathogens (e.g. *Legionella pneumophila* and *P. aeruginosa*) can enter a VBNC state in which they are not detectable using standard culture-based analysis but are still viable and retain their virulence (Mansi et al. 2014). Moreover, these VBNC microorganisms may regain their ability to be cultured under favourable conditions, which corresponds to the observed bacterial re-growth observed after 24 hours (culture-based analysis). Strauss et al. (2019) then applied Illumina next-generation sequencing coupled with EMA viability treatment to identify the primary pathogenic or opportunistic pathogenic genera, capable of surviving SODIS-CPC treatment in a 10.6 L CPC-reactor (Strauss et al. 2019). Results from the study indicated that intact and potentially viable bacterial cells belonging to 11 different bacterial genera (e.g. *Acinetobacter*, *Campylobacter*, *Legionella*, *Mycobacterium* and *Pseudomonas* amongst others) were detected in the SODIS-CPC treated tank water. Monitoring for the presence of VBNC microorganisms following water treatment is thus essential as these VBNC bacteria still pose a health risk as they are potentially infectious (Mansi et al. 2014).

While the survival of the *Cryptosporidium* oocysts after SODIS treatment using the solar reactor prototypes, may be attributed to the resilient nature of the oocyst wall (Hamilton et al. 2018), the ability of the opportunistic pathogenic bacteria (*Pseudomonas* spp., *Salmonella* spp., *Legionella* spp. and *Klebsiella* spp.) to survive large-scale solar-based disinfection strategies has been attributed to their ability to initiate various stress-response mechanisms and switch to a more tolerant phenotype upon exposure to environmental stressors, such as temperature and UV exposure (Jones, 1997; Fux et al. 2005). These stress-

responses may include the production of heat shock proteins and the initiation of DNA repair mechanisms, amongst others (Fields et al. 2002; Breidenstein et al. 2011). For example, Srivastava et al. (2008) indicated that the overexpression of the sigma factor *algT*, protects *Pseudomonas* spp. from heat stress and allows these organisms to persist during unfavourable conditions, while DNA repair mechanisms may be initiated in response to UV-induced DNA damage, through the activation of the SOS-regulon (upregulation of *recA* and *lexA*) or the photolyase enzyme (Zenoff et al. 2006). Similarly, Bojer et al. (2010) attributed the heat resistance of *K. pneumoniae* to the *clpK* genetic marker, which has been shown to correlate positively with thermotolerant phenotypes observed among clinical *Klebsiella* isolates. Microorganisms have also been reported to produce pigments or structures that may enable their survival under unfavourable conditions, as has been reported for *P. aeruginosa* where the production of pyocyanin has been hypothesised to protect *P. aeruginosa* from oxidative stress (inactivation mechanism of SODIS) (Hendiani et al. 2019). It is thus evident that microorganisms may employ numerous strategies to survive disinfection treatment and that additional treatment barriers may be required to reduce the survival of these target pathogens within water treatment systems. These strategies may include the addition of a cost-effective filtration system as a pre-treatment strategy to reduce microbial load entering the large-scale solar reactor prototypes (Hamilton et al. 2019).

3.3 Water safety plan and operational sustainability of the systems

As numerous factors may influence the quality of RHRW during the harvesting and/or treatment process, a WSP (Appendix B) for the utilisation of rainwater harvesting in combination with the large-scale solar reactor prototypes was developed. As the WSP was developed concurrently with the monitoring of the large-scale solar reactor prototypes during the field trials, the effectiveness of the various control measures was assessed by comparing site 1 with site 2, as these sites were located in two distinct settings that could be influenced by different anthropogenic activities and potential pollution sources as outlined in Appendix A.

447 The application of the WSP to characterise the risk associated with RHRW collected
448 at sites 1 and 2, indicated that the external hazards at site 1 (informal settlement) posed a
449 greater risk of contamination. The increased risk was primarily attributed to the influence of
450 potential pollution sources present near the catchment system (e.g. garbage disposal site,
451 surface run-off), tree branches obstructing a section of the conveyance system, organic debris
452 (e.g. dust/soil dispersed from the dirt pathway, leaves from the tree) within the conveyance
453 system and corrosion of the metal sheeting catchment system. Correspondingly, chemical and
454 microbial analysis of the untreated tank water samples collected from sites 1 and 2 revealed
455 that the untreated tank water collected from site 1 had higher levels of chemical contaminants
456 (e.g. cations) and microbial contaminants in comparison to site 2. For example, the
457 concentration of HPC was 0.72 log [3.50×10^5 CFU/100 mL (Tank 1) vs 6.90×10^4
458 CFU/100 mL (Tank 2-FF)] greater in the untreated tank water samples from site 1 (Tank 1), in
459 comparison to site 2 (Tank 2-FF).
460 The improved tank water quality at site 2 may also be attributed to the efficiency of the
461 implemented control measures at this site. The catchment surface at site 2 was painted with
462 a weather resistant roof paint that may have reduced the leaching of metal contaminants into
463 the collected tank water. Additionally, due to space availability a first-flush diverter was
464 connected between the catchment system and Tank 2-FF, which served as a control measure
465 to reduce the introduction of organic debris into the collection tank. However, the efficiency of
466 a first-flush diverter is dependent on the maintenance of the system, which entailed
467 cleaning/emptying the first-flush diverter after each rain event. The quality of RHRW collected
468 from site 1 may then be improved by removing the obstructing tree branches (source of
469 organic debris), implementing a regular gutter cleaning regime, installing a gutter screen at
470 the inlet of the RWH tank (due to space limitation a first-flush diverter could not be connected
471 to the current catchment system) and replacing the corroded metal sheeting on the catchment
472 system or painting the catchment system with a weather resistant roof paint.
473 As previously indicated, workshops were conducted with participating households
474 within the respective communities to outline the operational maintenance of the large-scale

475 solar reactor prototypes and rainwater harvesting systems (Fig. A3). Subsequent monitoring
476 of the operational sustainability of the solar reactor prototypes at both sites indicated that
477 system maintenance was limited to cleaning the surface of the PMMA reactor tubes (prevent
478 dust accumulation that will influence UV transmittance), with no system components needing
479 replacement during the study period. The robustness of system components therefore also
480 needs to be taken into consideration when designing water treatment systems for use in rural
481 areas and informal settlements, where replacement components may not be readily available.
482 During the study period, households who had access to the treated tank water (Prototype I
483 and II) at sites 1 (13 households) and site 2 (5 households), primarily reported using the treated
484 tank water for domestic activities such as cleaning of their homes, laundry and washing.
485 As noted by Mahmud et al. (2007), the aim of a WSP for small community water
486 supplies should be to achieve an overall and sustained reduction in microbial
487 contaminants/sanitary risks, rather than aim for the complete removal of microbial
488 contaminants. The WSP outlined in the current study thus serves to reduce the contamination
489 of RHRW by reducing "preventable contaminant entry" (e.g. organic debris and faecal matter
490 containing an increased microbial load from washing into the storage tank) into the storage
491 tank, whereafter treatment with the large-scale solar reactor prototypes may further reduce
492 the microbial contaminants to within drinking water standards.

493 **4. Conclusions**

494 The physico-chemical and chemical quality of the Tank 1 and 2-FF and Prototype I and II
495 treated rainwater samples adhered to the respective drinking water guidelines, with an
496 improvement in quality observed at site 2 where the first-flush diverter was installed. Lower
497 indicator bacterial counts were also recorded in the tank water samples collected from site 2
498 (Tank 2-FF and Prototype II) where the first-flush diverter was installed and fewer hazards
499 were identified that may influence the tank water quality (WSP), in comparison to site 1 (Tank
500 1 and Prototype I). The installation of a first-flush diverter system may thus serve as an
501 inexpensive pre-treatment strategy that may improve the overall quality of RHRW, while the

establishment of a WSP may aid in identifying potential hazards/hazardous events that may influence water safety.

Although both reactor prototypes were able to significantly improve the microbial quality of the tank water after an 8 hour solar treatment, HPC exceeding the DWAF (1996) drinking water guideline limit were recorded in 43% of the Prototype I treated samples. Nevertheless, a mean 1.01 log reduction in heterotrophic bacteria was recorded for these samples, which would decrease the health risk associated with using the treated rainwater (in comparison to the utilisation of untreated rainwater). Results from the EMA-qPCR and PMA-qPCR analysis indicated that *E. coli*, enterococci, *Klebsiella* spp., *Legionella* spp., *Pseudomonas* spp., *Salmonella* spp. and *Cryptosporidium* oocysts were reduced by 74.43% in both reactor prototypes. While molecular analysis indicated that the target organisms in the treated rainwater samples were not reduced to below the detection limit, based on national and international drinking water guidelines, the large-scale solar reactor prototypes used in the current study may effectively treat rainwater to within drinking water standards. The 88 L and 140 L solar reactor prototype treatment systems may thus provide a viable water provision solution for the inhabitants of rural areas and urban informal settlements in sub-Saharan Africa.

Conflicts of interests

The authors have no conflicts to declare.

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524 **References**

525 Alonso, J.L., Amorós, I., Guy, R.A., 2014. Quantification of viable *Giardia* cysts and
526 *Cryptosporidium* oocysts in wastewater using propidium monoazide quantitative real-time
527 PCR. Parasitol. Res. 113, 2671-2678. https://doi.org/10.1007/s00436-014-3922-9.

528 Baker, W.P., Leyva, K.J., Klotz, P., Wellington, R.R., 2003. Investigation of bacteriophage as
529 indicators of fecal pollution in urban water samples. Journal of the Arizona-Nevada Academy
530 of Science. 36, 1-4. https://www.jstor.org/stable/40022384.

531 Bojer, M.S., Struve, C., Ingmer, H., Hansen, D.S., Krogfelt, K.A., 2010. Heat resistance
532 mediated by a new plasmid encoded Clp ATPase, ClpK, as a possible novel mechanism for
533 nosocomial persistence of *Klebsiella pneumoniae*. PLoS ONE. 5, e15467.
534 https://doi.org/10.1371/journal.pone.0015467.

535 Breidenstein, E.B.M., De La Fuente-Núñez, C., Hancock, R.E.W., 2011. *Pseudomonas*
536 *aeruginosa*: all roads lead to resistance. Trends Microbiol. 19, 419-426.
537 https://doi.org/10.1016/j.tim.2011.04.005.

538 Chang, M., McBroom, M.W., Beasley, R.S., 2004. Roofing as a source of non-point water
539 pollution. J. Environ. Manage. 73, 307-315. https://doi.org/10.1016/j.jenvman.2004.06.014.

540 Clements, T.L., Reyneke, B., Strauss, A., Khan, W., 2019. Persistence of viable bacteria in
541 solar pasteurised harvested rainwater. Water Air Soil Poll. 230, 130.
542 http://dx.doi.org/10.1007/s11270-019-4184-z.

543 Department of Water Affairs and Forestry (DWAF)., 1996. *South African Water Quality*
544 *Guidelines* 2nd ed. Volume 1: Domestic Water Use. Pretoria, South Africa: CSIR Environmental
545 Services. ISBN 0-7988-5338-7.

546 Dobrowsky, P.H., Lombard, M., Cloete, W.J., Saayman, M., Cloete, T.E., Carstens, M., Khan,
547 S., Khan, W., 2015. Efficiency of microfiltration systems for the removal of bacterial and viral

548 contaminants from surface and rainwater. Water Air Soil Pollut. 226, 33.
549 https://doi.org/10.1007/s11270-015-2317-6.

550 Dos Santos, S., Adams, E.A., Neville, G., Wada, Y., De Sherbinin, A., Mullin Bernhardt, E.,
551 Adamo, S.B., 2017. Urban growth and water access in sub-Saharan Africa: progress,
552 challenges, and emerging research directions. Sci. Total Environ. 607-608, 497-508.
553 https://doi.org/10.1016/j.scitotenv.2017.06.157.

554 Emenike, C.P., Tenebe, I.T., Omole, D.O., Ngene, B.U., Oniemayin, B.I., Maxwell, O., Onoka,
555 B.I., 2017. Accessing safe drinking water in sub-Saharan Africa: Issues and challenges in
556 South-West Nigeria. Sustain. Cities Soc. 30, 263-272.
557 https://doi.org/10.1016/j.scs.2017.01.005.

558 Fields, B.S., Benson, R.F., Besser, R.E., 2002. *Legionella* and Legionnaires' disease: 25 years
559 of investigation. Clin. Microbiol. Rev. 15, 506-526. https://doi.org/10.1128/CMR.15.3.506–
560 526.2002.

561 Fittipaldi, M., Nocker, A., Codony, F., 2012. Progress in understanding preferential detection
562 of live cells using viability dyes in combination with DNA amplification. J. Microbiol. Methods.
563 91, 276-289. https://doi.org/10.1016/j.mimet.2012.08.007.

564 Fux, C.A., Costerton, J.W., Stewart, P.S., Stoodley, P., 2005. Survival strategies of infectious
565 biofilms. Trends Microbiol. 13, 34-40. https://doi.org/10.1016/j.tim.2004.11.010.

566 Gikas, G.D., Tsihrintzis, V.A., 2012. Assessment of water quality of first-flush roof runoff and
567 harvested rainwater. J. Hydrol. 466-467, 115-126.
568 https://doi.org/10.1016/j.jhydrol.2012.08.020.

569 Global Risks Report., 2019. *The Global Risks Report 2019 14th Edition*. Available:
570 http://www3.weforum.org/docs/WEF_Global_Risks_Report_2019.pdf. [2019, February 10].

571 Gwenzi, W., Nyamadzawo, G., 2014. Hydrological impacts of urbanization and urban roof
572 water harvesting in water-limited catchments: a review. Environ. Process. 1, 573-593.
573 https://doi.org/10.1007/s40710-014-0037-3.

574 Hamilton, K.A., Waso, M., Reyneke, B., Saeidi, N., Levine, A., Lalancette, C., Besner, M-C.,
575 Khan, W., Ahmed, W., 2018. *Cryptosporidium* and *Giardia* in wastewater and surface water
576 environments. J Environ Qual. 47, 1006-1023. https://doi.org/10.2134/jeq2018.04.0132.

577 Hamilton, K.A., Reyneke, B., Waso, M., Clements, T.L., Ndlovu, T., Khan, W., Digiovanni, K.,
578 Rakestraw, E., Montalto, F., Haas, C.N. Ahmed, W., 2019. A global review of the
579 microbiological quality and potential health risks associated with roof-harvested rainwater
580 tanks. Npj Clean Water. 2, 7. https://doi.org/10.1038/s41545-019-0030-5.

581 Hendiani, S., Pornour, M., Kashef, N., 2019. Quorum-sensing-regulated virulence factors in
582 *Pseudomonas aeruginosa* are affected by sub-lethal photodynamic inactivation. Photodiagn.
583 Photodyn. 26, 8-12. https://doi.org/10.1016/j.pdpdt.2019.02.010.

584 Jones, K., 1997. Strategies for survival. In: Coliforms and E. coli. Problem or solution? D. Kay
585 & C. Fricker, Eds. Cambridge, UK: The Royal Society of Chemistry.

586 Mahmud, S.G., Abu Jafar Shamsuddin, S., Feroze Ahmed, M., Davison, A., Deere, D.,
587 Howard, G., 2007. Development and implementation of water safety plans for small water
588 supplies in Bangladesh: benefits and lessons learned. J. Water Health. 5, 585-597.
589 https://doi.org/10.2166/wh.2007.045.

590 Mansi, A., Amori, I., Marchesi, I., Marcelloni, A.M., Proietto, A.R., Ferranti, G., Magini, V.,
591 Valeriani, F., Borella, P., 2014. *Legionella* spp. survival after different disinfection procedures:
592 Comparison between conventional culture, qPCR and EMA-qPCR. Microchem. J. 112, 65-69.
593 https://doi.org/10.1016/j.microc.2013.09.017.

McGuigan, K.G., Conroy, R.M., Mosler, H., Du Preez, M., Ubomba-Jaswa, E., Fernandez-Ibañez, P., 2012. Solar water disinfection (SODIS): a review from bench-top to roof-top. J. Hazard. Mater. 235-236, 29-46. https://doi.org/10.1016/j.jhazmat.2012.07.053.

Nelson, K.L., Boehm, A.B., Davies-Colley, R.J., Dodd, M.C., Kohn, T., Linden, K.G., Liu, Y., Maraccini, P.A., McNeill, K., Mitch, W.A., Nguyen, T.H., Parker, K.M., Rodriguez, R.A., Sassoubre, L.M., Silverman, A.I., Wigginton, K.R., Zepp, R.G., 2018. Sunlight-mediated inactivation of health-relevant microorganisms in water: a review of mechanisms and modelling approaches. Environ. Sci.: Proc. Imp. 20, 1089. https://doi.org/10.1039/c8em00047f.

NHMRC and NRMMC (ADWG)., 2011. *Australian drinking water guidelines Paper 6 National Water Quality Management Strategy*. National Health and Medical Research Council, National Resource Management Ministerial Council Commonwealth of Australia, Canberra.

Reyneke, B., Dobrowsky, P.H., Ndlovu, T., Khan, S., Khan, W., 2016. EMA-qPCR to monitor the efficiency of a closed-coupled solar pasteurization system in reducing *Legionella* contamination of roof-harvested rainwater. Sci. Total Environ. 553, 662-670. https://doi.org/10.1016/j.scitotenv.2016.02.108.

Reyneke, B., Ndlovu, T., Khan, S., Khan, W., 2017. Comparison of EMA-, PMA- and DNase qPCR for the determination of microbial cell viability. Appl. Microbiol. Biotechnol. 101, 7371-7383. https://doi.org/10.1007/s00253-017-8471-6.

Reyneke, B., Cloete, T.E., Khan, S., Khan, W., 2018. Rainwater harvesting solar pasteurization treatment systems for the provision of an alternative water source in peri-urban informal settlements. Environ. Sci: Water Res. Technol. 4, 291-302. https://doi.org/10.1039/c7ew00392g.

617 Sánchez, A.S., Cohim, E., Kalid, R.A., 2015. A review on physicochemical and microbiological
618 contamination of roof-harvested rainwater in urban areas. Sustainability of Water Quality and
619 Ecology. 6, 119-137. https://doi.org/10.1016/j.swaqe.2015.04.002.

620 South African Bureau of Standards (SABS). 2005. South African National Standards (SANS)
621 241: In *Drinking water quality management guide for water services authorities*. 6th ed.
622 Annexure 1. ISBN 0-626-17752-9.

623 Srivastava, S., Yadav, A., Seem, K., Mishra, S., Chaudhary, V., Nautiyal, C.S. 2008. Effect of
624 high temperature on *Pseudomonas putida* NBRI0987 biofilm formation and expression of
625 stress sigma factor RpoS. Curr. Microbiol. 56, 453-457. https://doi.org/10.1007/s00284-008-
626 9105-0.

627 Strauss A., Dobrowsky P.H., Ndlovu T., Reyneke B., Khan W., 2016. Comparative analysis of
628 solar pasteurization versus solar disinfection for the treatment of harvested rainwater. BMC
629 Microbiol. 16, 289. https://doi.org/10.1186/s12866-016-0909-y.

630 Strauss A., Reyneke B., Waso M., Khan W., 2018. Compound parabolic collector solar
631 disinfection system for the treatment of harvested rainwater. Environ. Sci.: Water Res.
632 Technol. 4, 976-991. https://doi.org/10.1039/C8EW00152A.

633 Strauss A., Reyneke B., Waso M., Ndlovu T., Brink C., Khan S., Khan W., 2019. EMA-
634 amplicon-based taxonomic characterisation of the viable bacterial community present in
635 untreated and SODIS treated roof-harvested rainwater. Environ. Sci.: Water Res. Technol. 5,
636 91-101. https://doi.org/10.1039/c8ew00613j.

637 Ubomba-Jaswa, E., Fernández-Ibáñez, P., Navntoft, C., Polo-López, M.I., McGuigan, K.,
638 2010. Investigating the microbial inactivation efficiency of a 25 L batch solar disinfection
639 (SODIS) reactor enhanced with a compound parabolic collector (CPC) for household use. J.
640 Chem. Tech. Biotech. 85, 1028-1037. https://doi.org/10.1002/jctb.2398.

641 Waso, M., Khan, S., Khan, W., 2018. Microbial source tracking markers associated with
642 domestic rainwater harvesting systems: correlation to indicator organisms. Environ. Res. 161,
643 446-455. https://doi.org/10.1016/j.envres.2017.11.043.

644 World Health Organisation (WHO)., 2004. *Guidelines for drinking-water quality*. Rev. 3rd ed.
645 World Health Organisation. Geneva, Switzerland: WHO Press. ISBN: 92-4-154638-7.

646 World Health Organisation (WHO)., 2011. *Guidelines for drinking-water quality*. Rev. 4th ed.
647 World Health Organisation. Geneva, Switzerland: WHO Press. ISBN: 978-92-4-154815-1.

648 World Health Organisation (WHO)., 2016. Results of round 1 of the WHO international scheme
649 to evaluate household water treatment technologies. World Health Organisation.
650 https://apps.who.int/iris/handle/10665/204284.

651 Zenoff, V.F., Siñeriz, F., Farías, M.E., 2006. Diverse responses to UV-B radiation and repair
652 mechanisms of bacteria isolated from high-altitude aquatic environments. Appl. Environ.
653 Microbiol. 72, 7857-7863. https://doi.org/10.1128/AEM.01333-06.

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Table 1 Frequency of detection and mean concentrations (CFU/100 mL) of indicator organisms and target bacterial pathogens in the tank water samples collected from sites 1 and 2.

Organism	Site 1		Site 2	
	Tank 1 (n = 15)	Prototype I (n = 15)	Tank 2-FF (n = 18)	Prototype II (n = 18)
E. coli	67% (6)	BDL	51% (3)	BDL
Total coliforms	100% (1.5×10^4)	27% (42)	100% (1.0×10^3)	11% (2)
Enterococci	20% (3.0×10^3)	BDL	28% (2.2×10^3)	BDL
Faecal coliforms	73% (1.2×10^4)	BDL	22% (1.1×10^3)	BDL
Heterotrophic bacteria	100% (3.5×10^5)	50% (1.8×10^4)	100% (6.9×10^4)	86% (6.5×10^3)
Klebsiella spp.	100% (1.9×10^4)	BDL	17% (8.0×10^2)	BDL
Pseudomonas spp.	ND	ND	ND	ND
Salmonella spp.	60% (6.3×10^3)	BDL	6% (1.0×10^3)	BDL
Coliphages (PFU/mL)	ND	ND	ND	ND

BDL – below detection limit; ND – not detected; PFU – plaque forming units

675 **Figure Legends:**

676 **Fig. 1. (A)** The Prototype I (140 L) solar reactor installed at Site 1. **(B)** The Prototype II (88 L)
677 solar reactor installed at Site 2. The red arrow indicates the first-flush diverter which was
678 connected to Tank 2-FF.

679 **Fig. 2.** Principle component analysis of the cations affecting the tank water quality for site 1
680 (Tank 1 and Prototype I) and 2 (Tank 2-FF and Prototype II). The directionality of the arrows
681 indicate the correlation (same = positive; opposite = negative) between the different variables
682 and illustrate the predominant variables best describing the collected tank water samples.

683 **Fig. 3.** Box and whiskers plot illustrating the distribution of the intact cells or oocysts/100 mL
684 recorded for each of the target organisms using EMA-qPCR (*E. coli*, enterococci, *Klebsiella*
685 spp., *Legionella* spp., *Pseudomonas* spp. and *Salmonella* spp.) and PMA-qPCR
686 (*Cryptosporidum* oocysts) in the tank water samples collected from **(A)** site 1 and **(B)** site 2.
687 The whiskers at the end of each box indicate the minimum and maximum values, while the
688 box is defined by the lower and upper quartiles and the mean value.